Figure 1

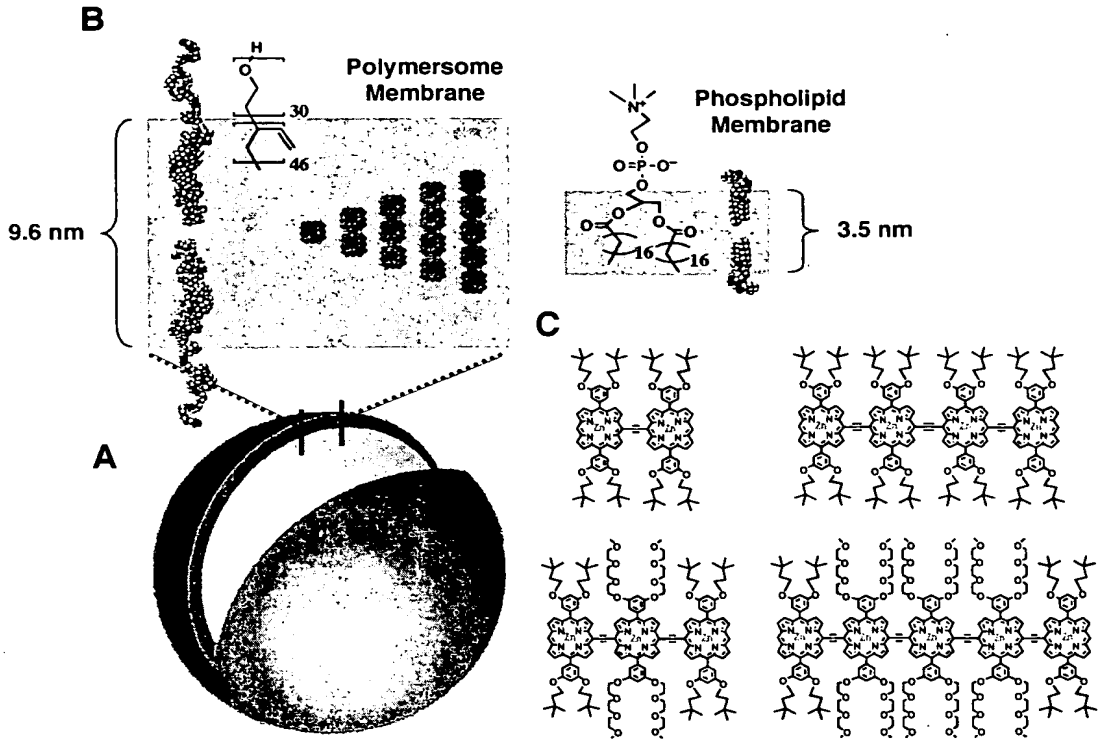

B
Polymersome Membrane
30
46
9.6 nm
Phospholipid Membrane
3.5 nm
16
16
A
C

Figure 2

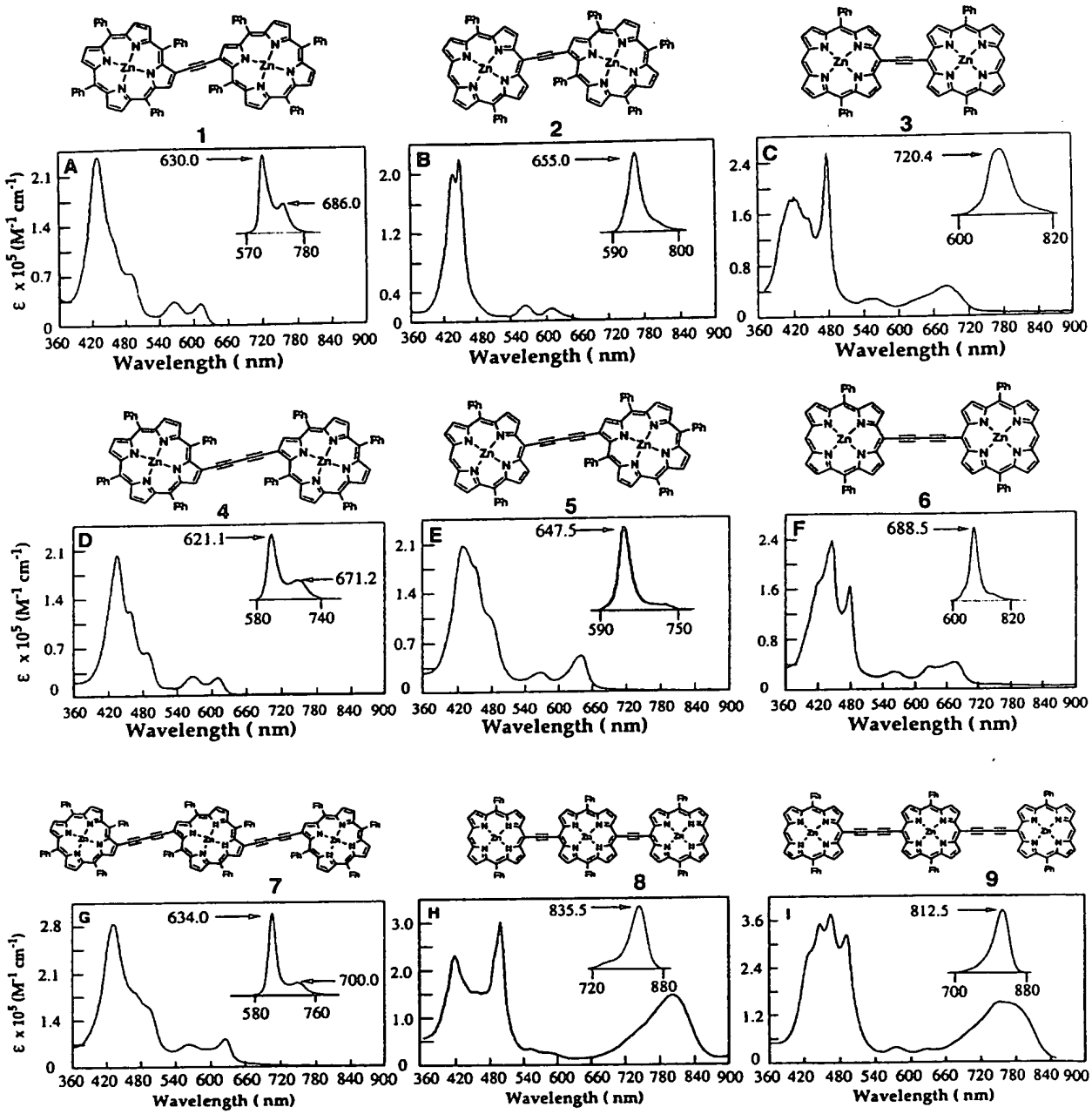

1
2
3
A
630.0
686.0
570
780
B
655.0
590
800
C
720.4
600
820
4
5
6
D
621.1
671.2
580
740
E
647.5
590
750
F
688.5
600
820
7
8
9
G
634.0
700.0
580
760
H
835.5
720
880
I
812.5
700
880
ε x 10^5 (M^-1 cm^-1)
Wavelength (nm)
360 420 480 540 600 660 720 780 840 900

Figure 3

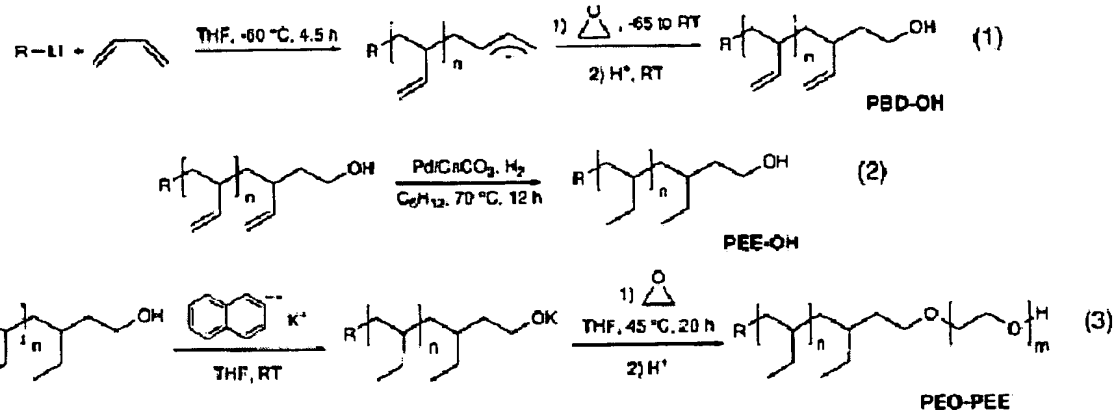

R–Li +
THF, -60 °C, 4.5 h
1) , -65 to RT
2) H+, RT
OH
(1)
PBD-OH
Pd/CaCO3, H2
C6H12, 70 °C, 12 h
(2)
PEE-OH
K+
THF, RT
OK
THF, 45 °C, 20 h
(3)
PEO-PEE

Figure 4

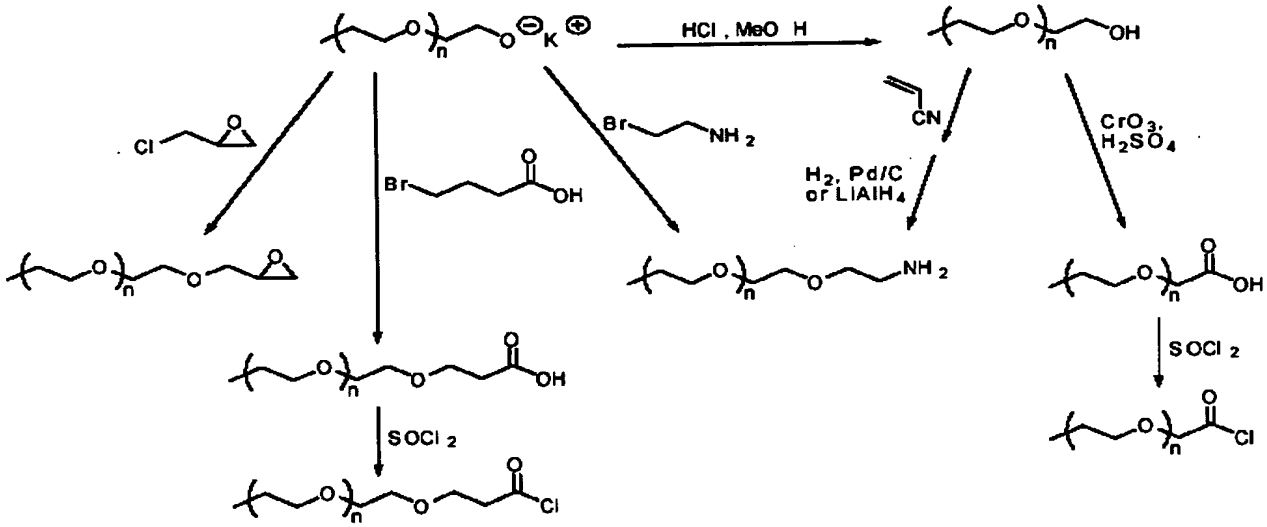

HCl , MeO H
Cl
Br
OH
Br
NH 2
CN
H2, Pd/C
or LiAlH4
CrO3,
H2SO4
NH 2
OH
SOCl 2
SOCl 2
Cl

Figure 5

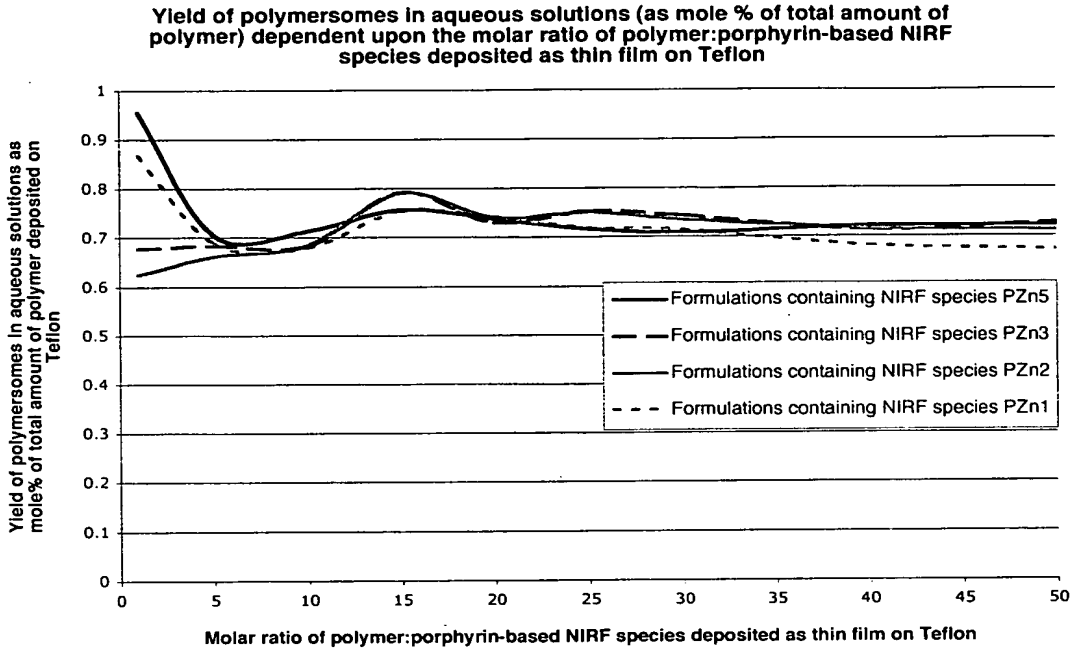

Yield of polymersomes in aqueous solutions (as mole % of total amount of polymer) dependent upon the molar ratio of polymer:porphyrin-based NIRF species deposited as thin film on Teflon
Yield of polymersomes in aqueous solutions as mole% of total amount of polymer deposited on Teflon
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
0
5
10
15
20
25
30
35
40
45
50
Formulations containing NIRF species PZn5
Formulations containing NIRF species PZn3
Formulations containing NIRF species PZn2
Formulations containing NIRF species PZn1
Molar ratio of polymer:porphyrin-based NIRF species deposited as thin film on Teflon

Figure 6

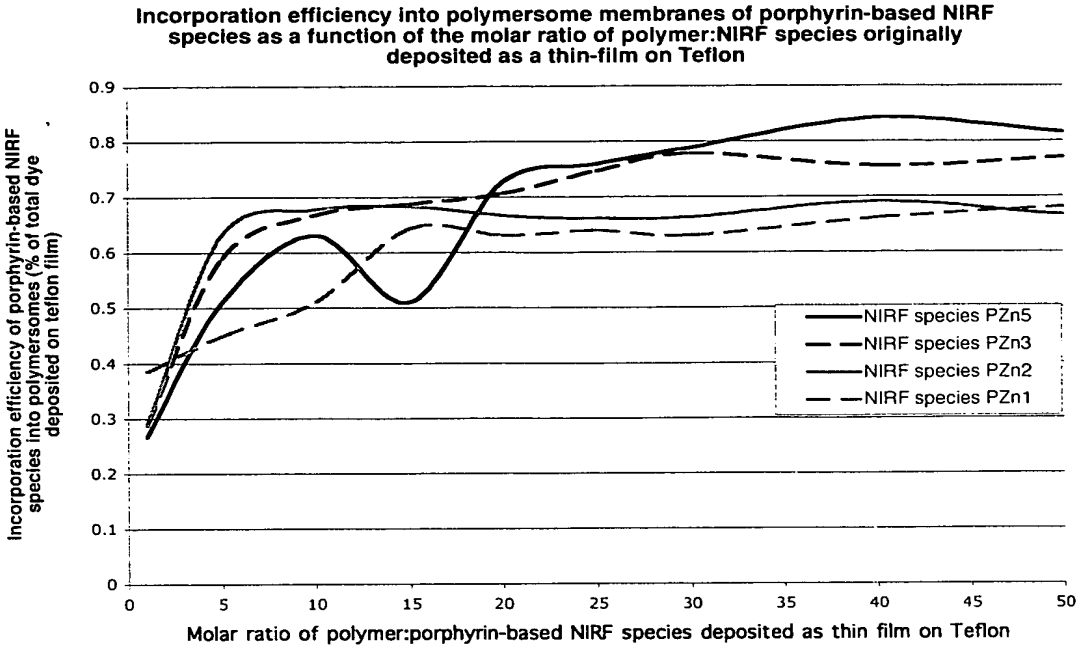

Incorporation efficiency into polymersome membranes of porphyrin-based NIRF species as a function of the molar ratio of polymer:NIRF species originally deposited as a thin-film on Teflon
Incorporation efficiency of porphyrin-based NIRF species into polymersomes (% of total dye deposited on teflon film)
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
0
5
10
15
20
25
30
35
40
45
50
Molar ratio of polymer:porphyrin-based NIRF species deposited as thin film on Teflon
NIRF species PZn5
NIRF species PZn3
NIRF species PZn2
NIRF species PZn1

Figure 7

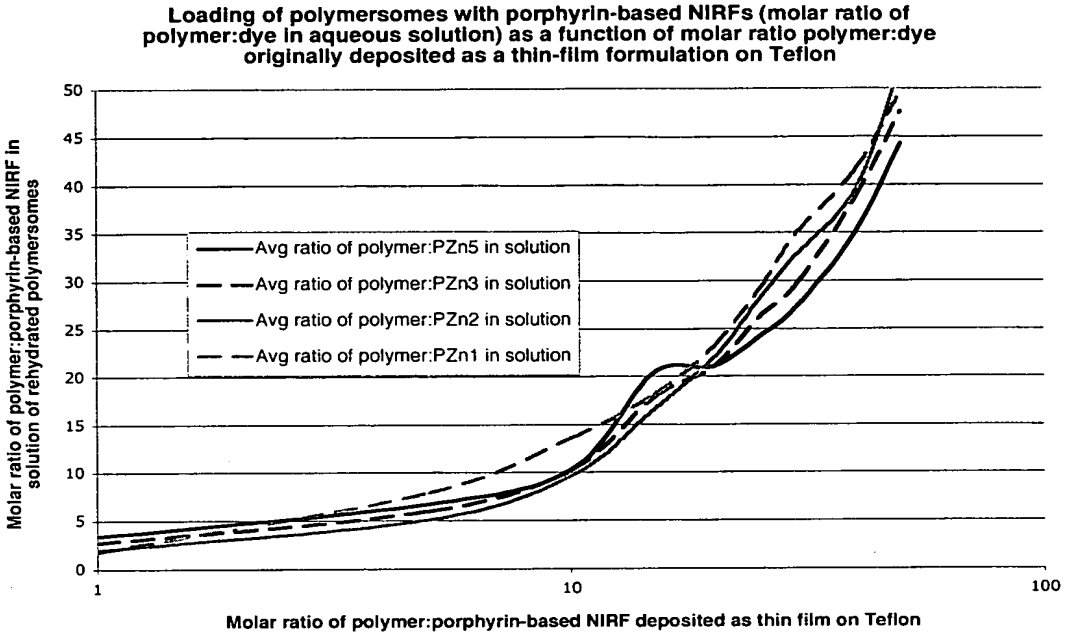

Loading of polymersomes with porphyrin-based NIRFs (molar ratio of polymer:dye in aqueous solution) as a function of molar ratio polymer:dye originally deposited as a thin-film formulation on Teflon
Molar ratio of polymer:porphyrin-based NIRF in solution of rehydrated polymersomes
50
45
40
35
30
25
20
15
10
5
0
1
10
100
Avg ratio of polymer:PZn5 in solution
Avg ratio of polymer:PZn3 in solution
Avg ratio of polymer:PZn2 in solution
Avg ratio of polymer:PZn1 in solution
Molar ratio of polymer:porphyrin-based NIRF deposited as thin film on Teflon

Figure 8

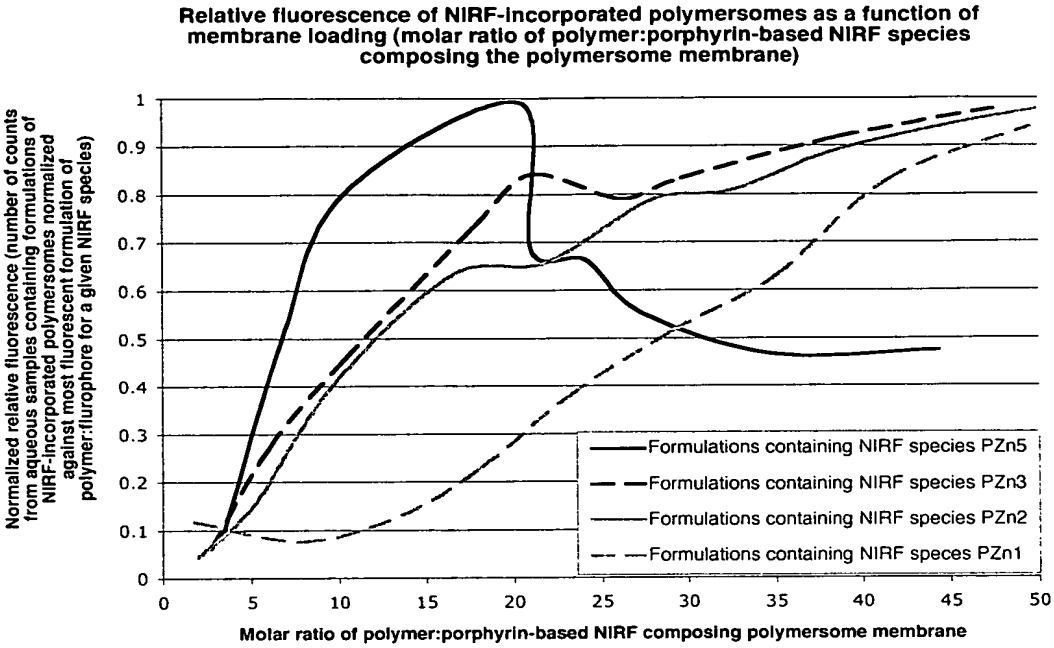

Relative fluorescence of NIRF-incorporated polymersomes as a function of membrane loading (molar ratio of polymer:porphyrin-based NIRF species composing the polymersome membrane)
Normalized relative fluorescence (number of counts from aqueous samples containing formulations of NIRF-incorporated polymersomes normalized against most fluorescent formulation of polymer:flurophore for a given NIRF species)
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
0
5
10
15
20
25
30
35
40
45
50
Molar ratio of polymer:porphyrin-based NIRF composing polymersome membrane
Formulations containing NIRF species PZn5
Formulations containing NIRF species PZn3
Formulations containing NIRF species PZn2
Formulations containing NIRF speces PZn1

Figure 9

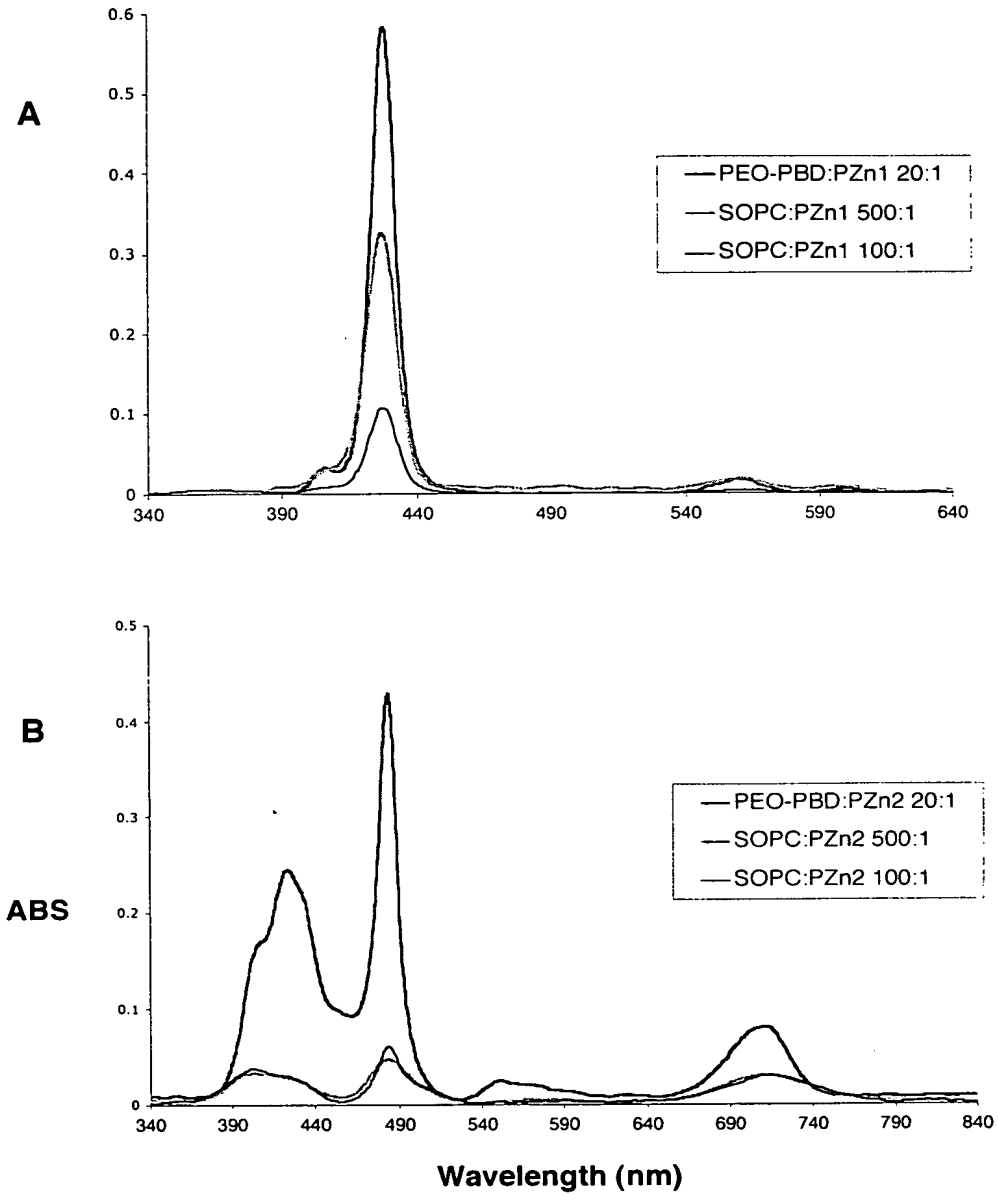
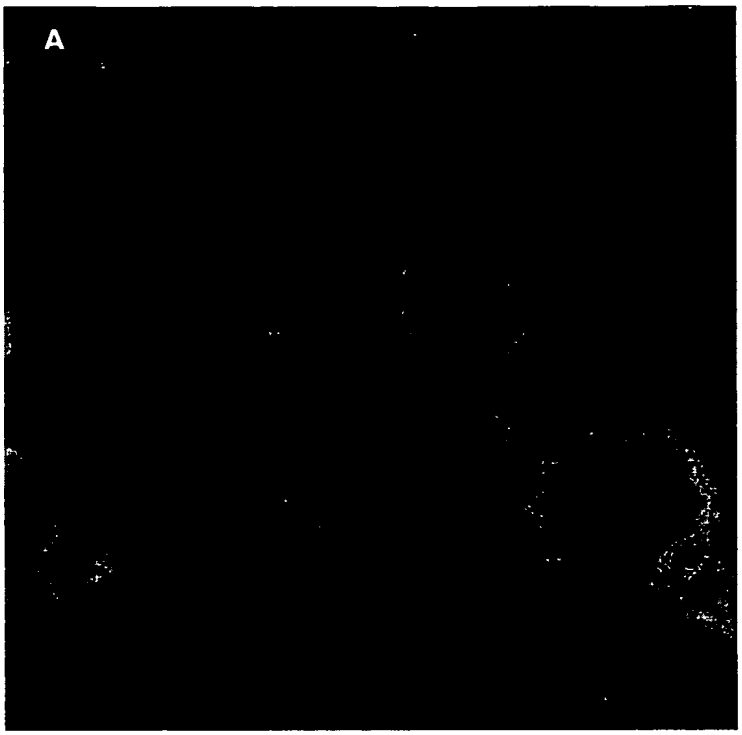

A
B
ABS
0.6
0.5
0.4
0.3
0.2
0.1
0
340
390
440
490
540
590
640
690
740
790
840
PEO-PBD:PZn1 20:1
SOPC:PZn1 500:1
SOPC:PZn1 100:1
PEO-PBD:PZn2 20:1
SOPC:PZn2 500:1
SOPC:PZn2 100:1
Wavelength (nm)

Figure 10



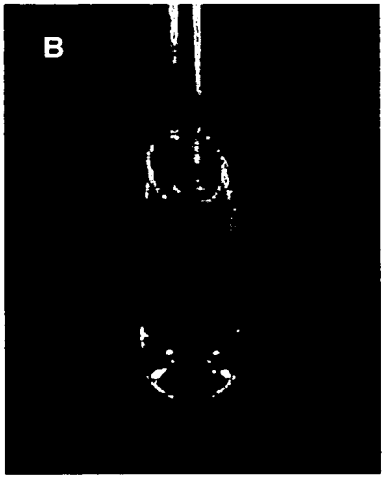


Figure 11

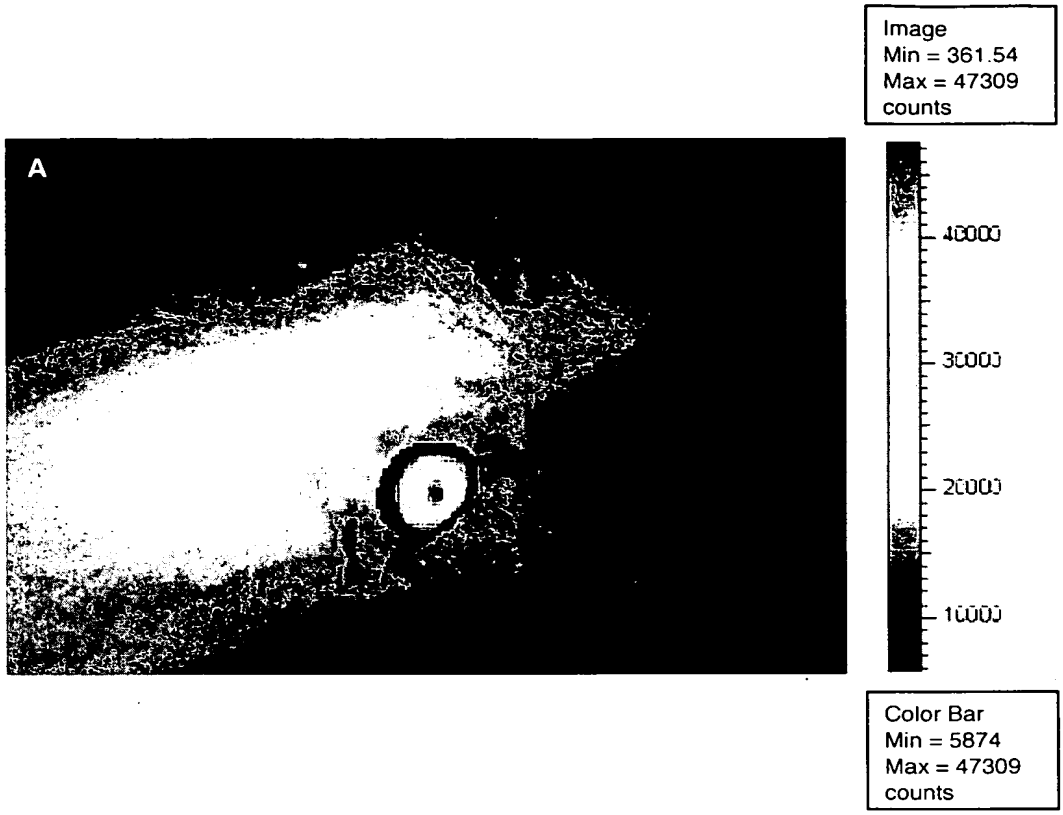

A
Image
Min = 361.54
Max = 47309
counts
40000
30000
20000
10000
Color Bar
Min = 5874
Max = 47309
counts
B
250
200
150
100
50
0

Figure 12

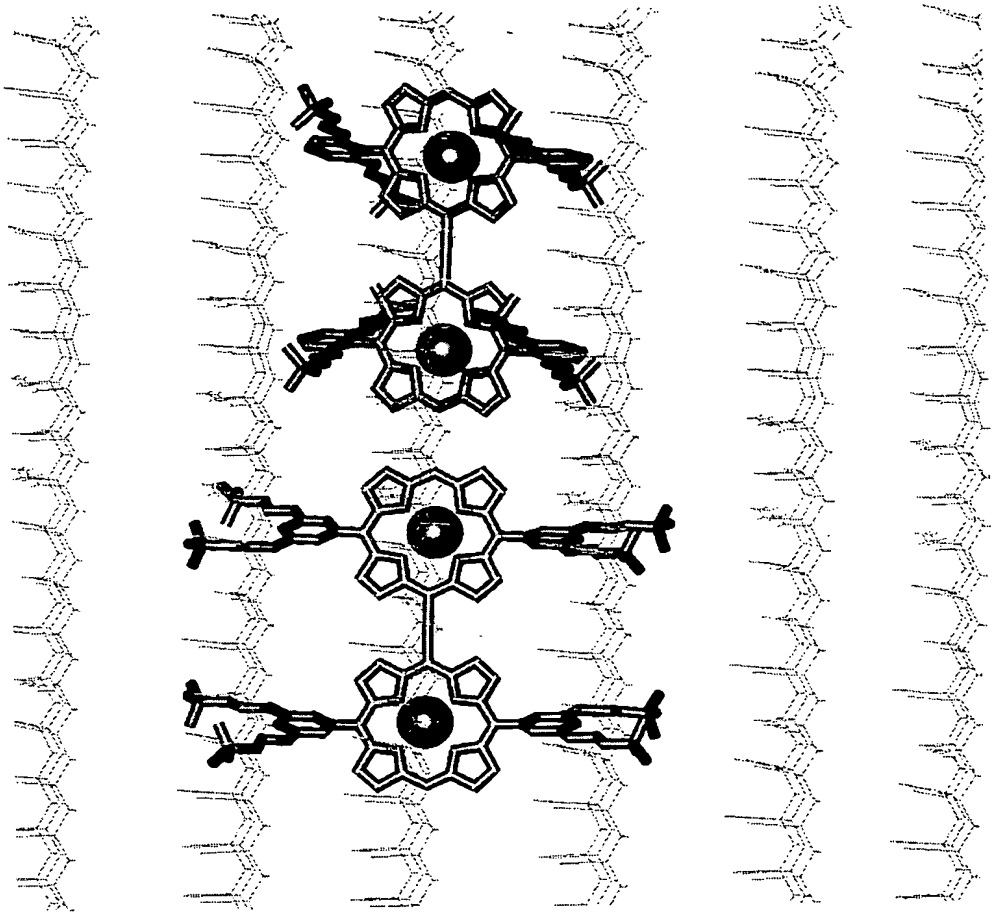
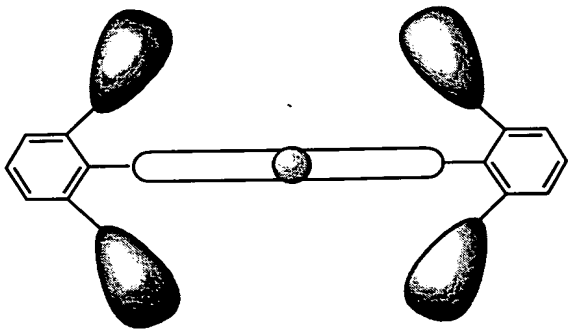
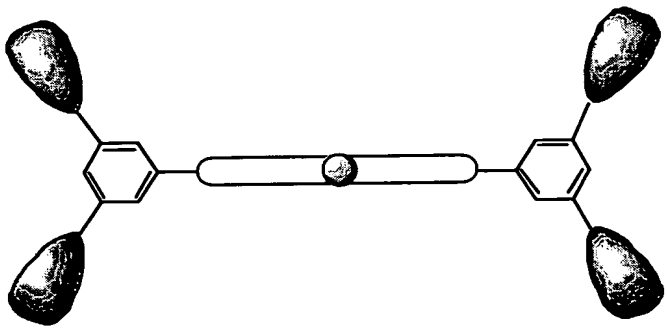


Figure 13

2, 6 substitution –



3, 5 substitution –



- 2,6 substituted oligomers experience blue-shift in fluorescence emission when incorporated in polymersomes as compared to free compound in THF
- 3,5 substituted oligomers experience red-shift in fluorescence emission when incorporated in polymersomes as compared to free compound in THF

Figure 14

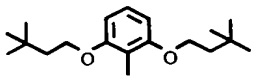
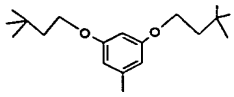
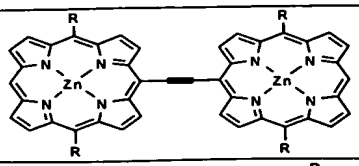
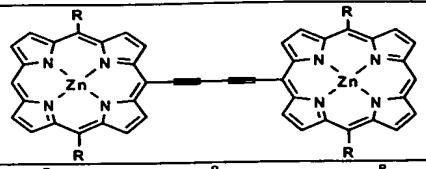
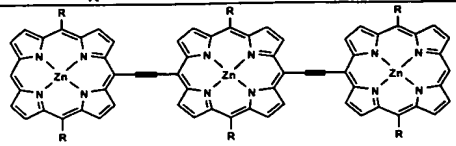
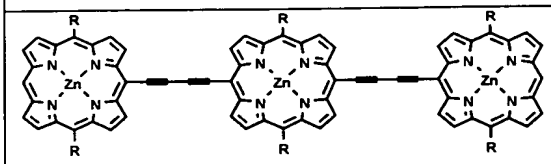
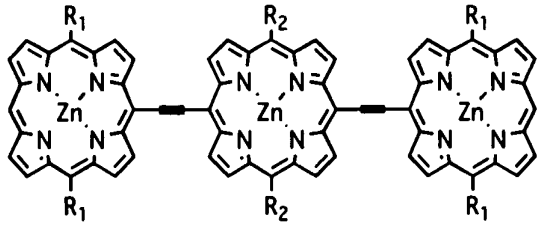
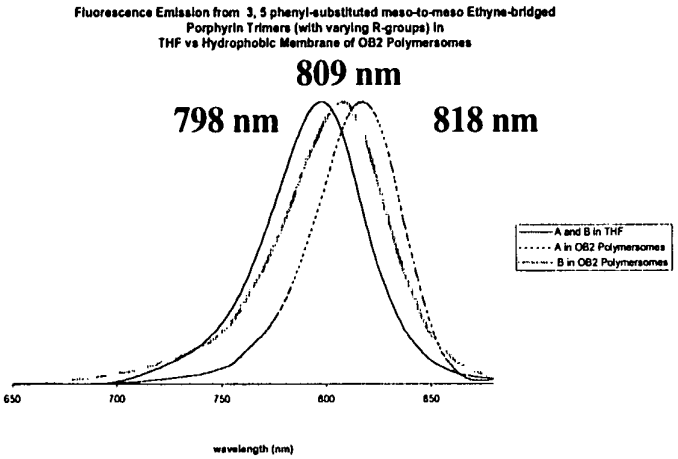
Compound	Emission in THF	R=  in polymersomes	R=  in polymersomes
	708 nm	-13 nm	+15 nm
	685 nm	-5 nm	+12 nm
	797 nm	-35 nm	+20 nm
	757	Same	Same

Figure 15

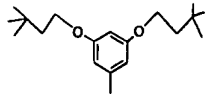
- Based on nature of R group off of phenyl ring



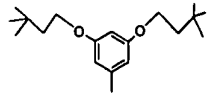
e.g. 3, 5 substituted meso-to-meso ethyne-bridged trimer



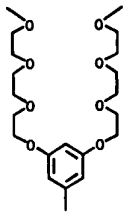
A: $R_1 = R_2 =$



B: $R_1 =$



$R_2 =$



Docket No.: UPN-4290
App No.: Not Yet Assigned
Filed: Herewith
Title: Polymersomes Incorporating Highly Emissive Probes
Inventors: Daniel A. Hammer, Michael J. Therien, Paiman Peter Ghoroghchian
Attorney: John A. Harrelson, Jr.
Phone: (215) 568-3100
Sheet 15 of 16

Figure 16